Claims:

1. Microorganism, characterized by the presence of a DNA sequence encoding a functional chaperone of a psychrophilic bacterium.

- 2. Microorganism according to claim 1, characterized in the DNA sequence encoding a functional chaperonin of a psychrophilic bacterium.
- 3. Microorganism according to claim 1, characterized in the DNA sequence encoding the chaperonin Cpn60 and/or Cpn 10 (SEQ ID No 1 and/or 2) of Oleispira antarctica.
- 4. Microorganism according to claim 1, characterized in the DNA sequence encoding a functional homolog of the chaperonin Cpn60 and/or Cpn10 of Oleispira antarctica (Seq ID Nr. 1 and/or 2) from a psychrophilic bacterium.
- 5. Microorganism according to claim 4, characterized in that the psychrophilic bacterium is selected from the group consisting of *Moraxella*, and *Alteromonas haloplanktis*.
- 6. Microorganism according to claim 1, characterized in the DNA sequence encoding a functional mutant of the chaperonin Cpn60 and/or Cpn 10 (Seq ID No 1 and/or 2) of Oleispira antarctica.
- 7. Microorganism according to claim 1, characterized in the DNA sequence encoding the stabilized single ring mutant chaperonin Glu461Ala/Ser463Ala/Val464Ala of Cpn60 (Seq ID No 11) or the mutant chaperonin Lys468Thr/Ser471Gly and/or Cpn 10.
- 8. Microorganism according to one of the preceding claims, which is selected among animal cell lines, plant cell lines, gram-positive or gram-negative bacteria, fungi and yeasts.
- Microorganism according to one of the preceding claims, characterized in that the heterologous protein has enzymatic activity or hormonal activity in its native conformation.
- 10. Microorganism according to one of the preceding claims, characterized in that the DNA sequence encoding a functional chaperone is located chromosomally, extrachromosomally, or mitochondrially, or in chloroplasts of plants.
- 11. Process for producing a protein by heterologous expression in a host microorganism containing a gene sequence encoding the heterologous protein, characterized in that a microorganism according to one of the preceding claims is used.
- 12. Process according to claim 11, characterized in that the host organism is cultivated at a temperature below 25 °C, preferably 4 to 15 °C.

13. Process according to claim 11 or 12, characterized in that the heterologous protein is selected from the group consisting of mammalian proteins, psychrophilic mammalian or bacterial proteins, mesophilic bacterial, fungal or yeast proteins, and mutant or fusion variants thereof.

- 14. Process for changing the conformation of denatured proteins into their native and/or active conformation, characterized by the step of contacting the denatured protein with a functional chaperone of a psychrophilic bacterium.
- 15. Process according to claim 14, characterized in that the chaperone is the chaperonin Cpn60 and/or Cpn 10 (Seq ID No 1 and/or 2) of *Oleispira antarctica* in presence of at least one nucleotide, preferably adenosine triphosphate.
- 16. Process according to claim 11, characterized in that the chaperone is a functional homolog of the chaperonin Cpn60 and/or Cpn 10 (Seq ID No 1 and/or 2) from a psychrophilic bacterium or a functional mutant of the chaperonin Cpn60 and/or Cpn 10 (Seq ID No 1 and/or 2) of Oleispira antarctica.
- 17. Process according to one of claims 11 to 16, characterized in that the contacting is performed extracellularly or *in vitro*.
- 18. Process according to claim 17, characterized in that the contacting uses at least one immobilized chaperone.
- 19. Plant, characterized in that it can grow at lower ambient temperatures due to the presence of a DNA sequence encoding a cold-active functional chaperone of a psychrophilic bacterium or plant.
- 20. Plant according to claim 19, characterized in the DNA sequence encoding a functional chaperonin selected from the group consisting of Cpn60 and/or Cpn 10 (SEQ ID No 1 and/or 2) of *Oleispira antarctica*, a functional homolog thereof, and the stabilized single ring mutant chaperonin Glu461Ala/Ser463Ala/Val464Ala of Cpn60 (Seq ID No 11).

Figure 1:

Amino acid sequences of Cpn60 and Cpn10:

SEQ ID No 1: Cpn10 (encoded by nucleotides pos. 458-751 of Figure 2):

MKIRPLHDRIVVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA VNEGDVVVFGKYSGONTIDIDGEELLILNESDIYGVLEA

SEO ID No 2: Cpn60 (encoded by nucleotides pos. 800-2446 of Figure 2):

MAAKDVLFGDSARAKMLVGVNILADAVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
AREIELKDKFENMGAQMVKEVASQANDQAGDGTTTATVLAQAIISEGLKSVAAGMN
PMDLKRGIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKV
GKEGVITVEEGKGLEDELDVVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKK
IDNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVAAVKAPGFGD
RRKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEAS
VNTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVD
DALHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQI
AGNAGAEGSVVVDKVKSGTGSFGFNASTGEYGDMIAMGILDPAKVTRSSLQAAASI
AGLMITTEAMVADAPVEEGAGGMPDMGGMGGMGGMPGMM

Figure 2:

SEO ID No 3: DNA coding for Cpn60 and Cpn10:

Cpn10, pos. 458-751

Cpn60, pos. 800-2446

atcaaaaatgcagcaaggacagattcctgcccaagaattagcagaaggtttcttgttagcactggccggcgctttattattaacgccgg
gttttgtcactgatgcgctgggttttacattactcgtccccgcgacgcgtaaagcgttggtccataaggtgattgcatttattacccctc
gcatgatgactgcaagcagctttcaagcgacgggtagttttcaggaaggctcgtttaaagatgtacattcgcacactgactcgcaaagca
gtcatgaaaaaatcacaattgaaggcgaatataccaaagacgataagtaggtatttttcggctagccgttgaaatcctagtaaaagccc

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cgataaattaaccatctatttttcacagaggcaatttagcctttgtttaccttattgatcctaatacttgggatccaacagttggagagtctagca a atgaa a at cegt ceatta cat gate gt att gt tege cegt a a a gaa gaa gaa gaa cege a act geg gg t gg tatt att tt taccel of the control of tgggcgctgcggcagaaaaaccaaatcaaggtgttgttatctctgtgggtactggccgtattcttgataatggttcagtgcaagcgctggc ggttaacgaaggcgatgttgtcgtttttggtaaatactcaggtcaaaatactatcgatatcgatggtgaagaattattgattttgaatga tattatttggtgatagcgcacgcgcaaaaatgttggtaggtgtaaacattttagccgacgcagtaagagttaccttaggacctaa aggtcgtaacgttgttatagaaaaatcatttggtgcaccgatcatcaccaaagatggtgtttctgttgcgcgtgaaatcgaattgaaagaca aattcgaaaacatgggcgcacagatggttaaggaagttgcttctcaagccaacgaccaagccggtgacggcacaacgacagcgact ccgatgaaacggttggtcgtttaattgctgaagcgatggaaaaagtcggtaaagaaggtgtgattaccgttgaagaaggcaaaggccttgaagacgagcttgatgttgtagaaggcatgcagttcgatcgcggttacttgtctccgtacttcatcaacaaccaagaaaaaatgaccgta gaaatggaaaatccattaattctattggttgataagaaaattgataaccttcaagagctgttgccaattcttgaaaacgtcgctaaatcaggt cgt ccattatt gateg ttg ctg aag at gttg aag ge caag cactag caa catt gg tag taaa caactt ge ge gg cacatt caag gtt ge ge gateg aag act ge gateg aan act ge gateg aag act ge gateg aag act ge gateg aag act ge gateg aan act ge gateg act ge gateg act ge gateg aan act ge gateg actagcggttaaagcccctggttttggcgatcgtcgtaaagcgatgttgcaagatcttgccatcttgacgggtggtcaggttatttctgaagag tggcgcaggtactgaagcaagcgttaatactcgtgttgaccagatccgtgctgaaatcgaaagctcgacttctgattacgacatcgaaaagttacaagaacgcgttgctaagcttgcgggcggcgttgccgtgattaaggttggtgcgggttctgaaatggaaatgaaagaagaagaa gaccgtgttgacgatgcacttcatgcaactcgcgcagcggttgaagaaggtgttgttgcgggtggtggtggttgctttgattcgcgcactctctt cag taaccgt tg ttg gt gataacgaa gat caaaacgt cg gt att gc att gg cact tcg tg cg at gg aag ctcct at ccg tcaaatcg cg at gatagaa gc tcct at ccg tcaaatcg cg at gatagaa gc tcct at ccg tcaaatcg cg at gatagaa gc tcct at ccg tcaaaacg tcg gt att gc att gg cact tcg tg cg at gg aag ctcct at ccg tcaaatcg cg at gatagaa gc tcct at ccg tcaaaacg tcg gt att gc att gg cact tcg tg cg at gg aag ctcct at ccg tcaaaacg tcg gt att gc att gg cact tcg tg cg at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt att gc att gg cact tcg tg cg at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg gt at gg aag ctcct at ccg tcaaaacg tcg gt at gg aag ctcct at ccg gt at gg at gat at gg at gat at gg at gg at gg at gat at gg at gggggtaacgcaggtgctgaagggtcagtggttgttgataaagtgaaatctggcacaggtagctttggttttaacgccagcacaggtgagtatggcgatatgattgcgatgggtattttagaccctgcaaaagtcacgcgttcatctctacaagccgcggcgtctatcgcaggtttgatgat cacaaccgaagccatggttgcggatgcgcctgttgaagaaggcgctggtggtatgcctgatatgggcggcatgggtggaatgggcg gtatgcctggcatgatgtaatcactttgtgattcattgtcctgatctgcttaccgtgtaaaaagatcaggctcaaggctgtctctataaaaag ttat gtaact aget gg cetata at gtt gagt te ctet gg gt gg cat gat et cat gg ta ctt cat gta ctt cat gag te ctt cat gg ta ctt cat

Figure 3:

SEQ ID No 4: Amino acid sequence of esterase cloned from Oleispira antarctica (EstRB8):

EstRB8 (encoded by nucleotides 1145 to 2143 Frame 2 of Figur 4) 333 aa

MKNTLKSSSRFSLKQLGTGALIISSLFFGGCTTTQQDNLYTGVMSLARDSAGLEVKTA SAGDVNLTYMERQGSDKDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG HGDSEQLLTTDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGGAISAIYSLSHPEKVKSL TLIDAAGVDGDTESEYYKVLAEGKNPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL RKTLARAEINNKIFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD VSAAAAFKKIIPQATVHIFPEVGHLPMVEIPSESAKVYEEFLSSIK

Figure 4:

SEQ ID No 5: DNA fragment from plasmid pBK1Est coding for esterase of *Oleispira* antarctica (EstRB8):

Nucleotide positions 1-100 correspond to reverse complement of positions 1196-1121 and 3799-3939 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene).

Positions 101-105 are *BamHI* – *Sau*3A1 fusion and positions 3795-3798 are *Sau*3A1-*Bam*HI-fusion.

gcgggacatggcgattcagaacaattattaacgactgattacggtctcataaaacaagccgagcgtttagatatcttcttatctggcttagg ggtta act cattte a categoeggta attea at gggggggggtate agegea at eta cagttt gagte acceagaga a agtta a a agtet the second control of the second control ofacattgatcgatgcagcaggtgtcgatggcgatactgaaagcgaatactacaaagttttggcagaaggtaagaatcctttaattgcaact gatgaagcaagttttgaataccgcatgggtttcaccatgactcagcctcctttcctaccttggccactaagaccttctttattacgtaaaacg ctagcccgtgccgagatcaataacaaaattttttccgatatgctgaaaaccaaagaacgtttaggaatgactaactttcaacagaaaattg aagtgaaaatggetcaacatecattgecaacactgattatgtggggcaaagaagategegttettgaegtateegeageageggeette ccaaattattcaacgaccaagctctgcggtaaaatcgcagtgggtttcttgttttcatcaacagcaacaaacgtgaaataccccgtaatcg catttttctgattatcaaaatacatactttccaccagcatattaacttcaacttttaaactcgtccgccctacctctataacactggcagtcaattcgacaatggtacctgcgggaacaggatgcttaaaatcgattcgatcactgctgacggttacgatgctttgtcgagaaaaacgagtcgct ataataaatagttaacagtatattgaactgagggtctgaagaactctaatacctctgaagaactttgaggccgctagagagaaaagacca atatttcatatatatatttcacactacccttatctcactagacttcccgcgcataggcgcaaacaatcaacgcaagttcacaataaagcggttc gctgcaacacatgccctagcgtctaaagtagcacgcacaacactggccagtcgtactagcccctttgcgattcgtgcagacgagcaac aageget attaaacttacctaaattte taaccaccaccattggt tett tte cacaaaact caaaaaact cgt caaatte cgettge aatttaaacgatatgcaagcggcggcggaagagctgcctttgatcgatcaagaagaaggagcagcaaaagaggaaaacaatcaaaaagaggaga a act tagge at tea a attacaga a att tagge at tatat cact gg caca at ge cag tage at tagge at tagg at taattcatggcggctcaaatgtattgctggcagaaacactgggcagcatggcagctaactgctgtattaatttgtctcaagaatattgtgttggccaagaaattaacgccaaccacatacgcggtgttcgttccggcatagtgactggcacagcaacgctagtacacaaaggaagaacctcccagatttgggaaattcgcatcgttaacgatccaaagaattcaaaaagcttctcgagagtacttctagagcggccgcggggcccatcgatt $t\^t c c a c c g g t g g g t a c c a g t a a g t g t a c c c a a t t c g c c t a t a g t g a g t c g t a t t a c a g t c g t t t a c a g t c g t t t a c a g t c g t t t a c a g t c g t a c a g t c g t a c a g t c g t a c a g t c g t a c a g t c g t a c a g t$

Figure 5:

Amino acid sequences expressed from vector pBK1CpnEst: - the co-expression of fragments encoding native chaperonines with the esterase gene (EstRB8), all from Oleispira antarctica

SEQ ID No 6: cpn10 (nucleotides 113 to 403: Frame 2 of Figure 6) 97 aa:

MKIRPLHDRIVVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA VNEGDVVVFGKYSGQNTIDIDGEELLILNESDIYGVLEA

SEQ ID No 7: cpn60 (nucleotides 455 to 2098: Frame 2 of Figure 6) 548 aa:

MAAKDVLFGDSARAKMLVGVNILADAVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
AREIELKDKFENMGAQMVKEVASQANDQAGDGTTTATVLAQAIISEGLKSVAAGMN
PMDLKRGIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKV
GKEGVITVEEGKGLEDELDVVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKK
IDNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVAAVKAPGFGD
RRKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEAS
VNTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVD
DALHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQI
AGNAGAEGSVVVDKVKSGTGSFGFNASTGEYGDMIAMGILDPAKVTRSSLQAAASI
AGLMITTEAMVADAPVEEGAGGMPDMGGMGGMGGMPGMM

SEQ ID No 8: estRB8 (nucleotides 2579 to 3577: Frame 2 of Figure 6) 333 aa:

MKNTLKSSSRFSLKQLGTGALIISSLFFGGCTTTQQDNLYTGVMSLARDSAGLEVKTA SAGDVNLTYMERQGSDKDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG HGDSEQLLTTDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGGAISAIYSLSHPEKVKSL TLIDAAGVDGDTESEYYKVLAEGKNPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL RKTLARAEINNKIFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD VSAAAAFKKIIPQATVHIFPEVGHLPMVEIPSESAKVYEEFLSSIK

Figure 6:

SEQ ID No 9: pBK1CpnEst: - the fusion of native chaperonine-coding fragments with esterase of Oleispira antarctica (EstRB8)

The DNA fragment coding for Cpn10 and Cpn60 is flanked by SacI site (pos. 69-75) and SalI site (encoded by pos. 2138-2143 of Figure 7):

Nucleotide positions 1-75 correspond to reverse complement of positions 1196-1121 and positions 5233-5273 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene)

Small letters — the Cpn10-Cpn60 encoding fragment,

Capital italics — fragments of vector pBK-CMV

Capital letters — fragment coding for EstRB8 from plasmid pBK1Est

ACAGGAAACAGCTATGACCTTGATTACGCCAAGCTCGAAATTAACCCTCACTAAAGGGA ACAAAAGCTGGAGCTCctaatacttgggatccaacagttggagagtctagcaaatgaaaatccgtccattacatgatcgtatt gttgttcgccgtaaagaagaagaagaccgcaactgcgggtggtattattttaccgggcgctgcggcagaaaaaccaaatcaaggtgttgt cttttttatttaacctacaaaatttaaggaaagatcatggctgctaaagacgtattatttggtgatagcgcacgcgcaaaaatgttggtaggt gtaaacattttagccgacgcagtaagagttaccttaggacctaaaggtcgtaacgttgttatagaaaaatcatttggtgcaccgatcatcac agccaacgaccaagccggtgacggcacaacgacagcgactgtactagcacaggcgattatcagcgaaggcttgaaatctgttgcgg gatacaaaagcaatcgctcaggtagggacaatctctgccaatgccgatgaaacggttggtcgtttaattgctgaagcgatggaaaaagt cggtaaagaaggtgtgattaccgttgaagaaggcaaaggccttgaagacgagcttgatgttgtagaaggcatgcagttcgatcgcggttacttgtctccgtacttcatcaacaaccaagaaaaaatgaccgtagaaatggaaaatccattaattctattggttgataagaaaattgataac cttcaagagctgttgccaattcttgaaaacgtcgctaaatcaggtcgtccattattgatcgttgctgaagatgttgaaggccaagcactagc aacattggtagtaaacaacttgcgcggcacattcaaggttgcagcggttaaagcccctggttttggcgatcgtcgtaaagcgatgttgca agatcttgccatcttgacgggtggtcaggttatttctgaagagctagggatgtctttagaaactgcggatccttcttctttgggtacggcaa

gttggtgcgggttctgaaatggaaatgaaagaaagaacgtgttgacgatgcacttcatgcaactcgcgcagcggttgaagaag gtgttgttgcgggtggtggtggttgctttgattcgcgcactctcttcagtaaccgttgttggtgataacgaagatcaaaacgtcggtattgcattgg cact tcg tgcg atgg aag ctcct at ccg tcaa atcgcg gg taacg cag gtgctg aag gg tcag tgg ttg ttg at aaag tgaa atctgcg transfer at the contract of the contracgcacaggtagetttggttttaacgccagcacaggtgagtatggcgatatgattgcgatgggtattttagaccctgcaaaagtcacgcgttcatctctacaagccgcggcgtctatcgcaggtttgatgatcacaaccgaagccatggttgcggatgcgcctgttgaagaaggcgctggtg gtatgcctgatatgggcggcatggggggtatggcggtatgcctggcatgatgtaatcactttgtgattcattgtcctgatctgcttaccgt CAAACACCAATACCAATCGCAAAAACTCATAAAACTAGCCGATCACCAAATCCC AAAAGCGTTCAAAAATGAAACGAGCACGTCACACAAAATCAATTTATACGCTAA CGAACCAGGTCAAACTTATCGTTTTTTGAGCACGTTTGTTCCACTAATGAAAGA GAAAAGTCGTTAATTCACTGGCTTTTGGCGTATCCGCACCTTCACATAGAAATTA GTAATGGCATGCTACTGGCCTTTAAAAAGAATCAGTTAATTGAAGAAACCTCGCT TATCTCAGCCATTACCGCTGTAGCCGAATTTGCGCTTATCCTCAGCCATGATTAAA CTGACGCCAATTAATAAGACATACTAATTAATAACTCCCTTAATTGAGAAGAA TAATGAAAAACACACTCAAATCCTCATCACGTTTTAGTCTGAAACAACTCGGCAC CGGCGCTCTGATTATCTCCAGTTTGTTCTTCGGTGGTTGCACCACAACACACAACAAG ATAATTTATACACAGGGGTTATGTCTCTTGCGAGAGACAGCGCTGGCCTAGAAGT TAAAACAGCCTCTGCCGGTGACGTCAATCTTACTTATATGGAACGCCAAGGCAGT GACAAGATAATGCCGAAAGCGTTATTTATTACACGGTTTCTCTGCTGATAAAG ATAACTGGATTCTTTTTACCAAAGAATTCGATGAAAAATATCATGTTATCGCTGTC GATTTAGCGGGACATGGCGATTCAGAACAATTATTAACGACTGATTACGGTCTCA TAAAACAAGCCGAGCGTTTAGATATCTTCTTATCTGGCTTAGGGGTTAACTCATTT CACATCGCCGGTAATTCAATGGGGGGGGCTATCAGCGCAATCTACAGTTTGAGTC ACCCAGAGAAAGTTAAAAGTCTTACATTGATCGATGCAGCAGGTGTCGATGGCG ATACTGAAAGCGAATACTACAAAGTTTTGGCAGAAGGTAAGAATCCTTTAATTGC AACTGATGAAGCAAGTTTTGAATACCGCATGGGTTTCACCATGACTCAGCCTCCT TTCCTACCTTGGCCACTAAGACCTTCTTTATTACGTAAAACGCTAGCCCGTGCCGA GATCAATAACAAAATTTTTTCCGATATGCTGAAAACCAAAGAACGTTTAGGAATG ACTAACTTTCAACAGAAAATTGAAGTGAAAATGGCTCAACATCCATTGCCAACAC TGATTATGTGGGGCAAAGAAGATCGCGTTCTTGACGTATCCGCAGCAGCGGCCTT CAAAAAAATAATTCCACAAGCAACTGTTCATATTTTTCCTGAAGTAGGCCACCTA CCTATGGTAGAAATTCCTAGTGAAAGCGCTAAAGTTTATGAAGAGTTTTTGTCCT CTATTAAATAAGAGCACATAATCATGACTGACTTATAAACAGCCAAGCATTTAAA ATGCTTGGCTGTTTATTTTAATGGCCAAATTATTCAACGACCAAGCTCTGCGGTAA

AATCGCAGTGGGTTTCTTGTTTTCATCAACAGCAACAAACGTGAAATACCCCGTA ATCGCATTTTTCTGATTATCAAAATACATACTTTCCACCAGCATATTAACTTCAAC TTTTAAACTCGTCCGCCCTACCTCTATAACACTGGCAGTCAATTCGACAATGGTAC CTGCGGGAACAGGATGCTTAAAATCGATTCGATCACTGCTGACGGTTACGATGCT GCAGTGCCACCGAATAACGTATCATGATGATTTGTTGTCTCTGGAAATACCGCTTT AGAAATAGTGGTTTTTGATACGCGCTTTCGCTGCGCAATAATATCTTCTCTGCTAA ACAGTATATTGAACTGAGGGTCTGAAGAACTCTAATACCTCTGAAGAACTTTGAG GCCGCTAGAGAGAAAAGACCAGTGATAATATTTCATCTTGCCATGAGAGCTTATC ATGAAAGCCTGTGCTTAAAATCAATCATTATATTTATTCATCTTTAATTGAAATAA TACCAATATATTCATATATATTTCACACTACCCTTATCTCACTAGACTTCCCGC GCATAGGCGCAAACAATCAACGCAAGTTCACAATAAAGCGGTTCGCTGCAACAC ATGCCCTAGCGTCTAAAGTAGCACGCACAACACTGGCCAGTCGTACTAGCCCCTT TGCGATTCGTGCAGACGAGCAACAAGCGCTATTAAACTTACCTAAATTTCTAACC ACCACCATTGGTTCTTTTCCACAAACTCAAAAAACTCGTCAAATCCGCTTGCAATT TAAACGCGATGACATAGATCTAATCGATTATCAAACCCGCATTCAAGCGCTCATT AAAAACGCACCACTGGCAAGAAGTTCTACCTGCACTGACCAATATGCAAGCGGC GGCGGAAGAGCTGCCTTTGATCGATCAAGAAGAAGGGAGCAGCAAAGAGGAAA ACAATCAAAAAGAGGAGAGCAATCAAATAAAAACGAGTTATTGAGGATTTTAAT TTTAAAACAGGTATATTAATACCCTCTCTCGTAGTAAACAATGACTGTATTTACAC AAAAATAAATAGAGGTATACCATGTCAAACATCTGGTTTGAAGTACCAAAGATTG AAGTATTAAACCGTCAAATGGAAAATACTGCCTGCAGCAACTTAGGCATTCAAAT TACAGAAATTGGCGATGATTATATCACTGGCACAATGCCAGCAGATGCACGTACC TTCCAGCCAATGGGACTGATTCATGGCGGCTCAAATGTATTGCTGGCAGAAACAC TGGGCAGCATGGCAGCTAACTGCTGTATTAATTTGTCTCAAGAATATTGTGTTGG CCAAGAAATTAACGCCAACCACATACGCGGTGTTCGTTCCGGCATAGTGACTGGC ACAGCAACGCTAGTACACAAAGGAAGAACCTCCCAGATTTGGGAAATTCGCATC GTTAACGATCCAAAGAATTCAAAAAGCTTCTCGAGAGTACTTCTAGAGCGGCCGCGGG CCCATCGATTTTCCACCCGGGTGGGGTACCAGGTAAGTGTACCCAATTCGCCCTATAGT GAGTCGTATTACAATTCACTGGCCGTCGTTTTAC

Figure 7:

Amino acid sequences expressed from vector pBK1CpnSREst: - the co-expression of the stabilized single ring mutant chaperonin with the esterase gene (EstRB8) from *Oleispira* antarctica (cpn10::stabilized single ring mutant Glu461Ala/Ser463Ala/Val464Ala::est)

SEQ ID No 10: cpn10 (nucleotides 113 to 403: Frame 2 of Figure 8) 97 aa:

MKIRPLHDRIVVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA VNEGDVVVFGKYSGQNTIDIDGEELLILNESDIYGVLEA

Below - Capital bold letters are the mutations introduced

SEQ ID No 11: stabilized single ring mutant of cpn60 (nucleotides 455 to 2098: Frame 2 of Figure 8) 548 aa:

MAAKDVLFGDSARAKMLVGVNILADAVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
AREIELKDKFENMGAQMVKEVASQANDQAGDGTTTATVLAQAIISEGLKSVAAGMN
PMDLKRGIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKV
GKEGVITVEEGKGLEDELDVVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKK
IDNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVAAVKAPGFGD
RRKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEAS
VNTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVD
DALHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQI
AGNAGAAGAAVVDKVKSGTGSFGFNASTGEYGDMIAMGILDPAKVTRSSLQAAASI
AGLMITTEAMVADAPVEEGAGGMPDMGGMGGMGGMPGMM

SEO ID No 12: EstRB8 (nucleotides 2579 to 3577: Frame 2 of Figure 8) 333 aa:

MKNTLKSSSRFSLKQLGTGALIISSLFFGGCTTTQQDNLYTGVMSLARDSAGLEVKTA SAGDVNLTYMERQGSDKDNAESVILLHGFSADKDNWILFTKEFDEKYHVIAVDLAG HGDSEQLLTTDYGLIKQAERLDIFLSGLGVNSFHIAGNSMGGAISAIYSLSHPEKVKSL TLIDAAGVDGDTESEYYKVLAEGKNPLIATDEASFEYRMGFTMTQPPFLPWPLRPSLL

RKTLARAEINNKIFSDMLKTKERLGMTNFQQKIEVKMAQHPLPTLIMWGKEDRVLD VSAAAAFKKIIPQATVHIFPEVGHLPMVEIPSESAKVYEEFLSSIK

Figure 8:

SEQ ID No 13: DNA sequence of vector pBK1CpnSREst: the expression cassette for the coexpression of the stabilized single ring mutant chaperonin with the esterase gene (EstRB8) from *Oleispira antarctica* (cpn10::stabilized single ring mutant Glu461Ala/Ser463Ala/Val464Ala::est)

Nucleotide positions 1-75 correspond to reverse complement of positions 1196-1121 and positions 5233-5273 correspond to reverse complement of 1043-952 of pBK-CMV vector (Stratagene)

DNA fragment coding for Cpn10 and Cpn60 is flanked by SacI site (pos. 69-75) and SalI site (pos. 2138-2143).

In the DNA sequence:

Small letters – the Cpn10-Cpn60 coding fragment,

Capital italics – fragments of vector

Capital letters – fragment coding for EstRB8 from plasmid pBK1Est

Capital bold letters = introduced mutations

gatacaaaagcaatcgctcaggtagggacaatctctgccaatgccgatgaaacggttggtcgtttaattgctgaagcgatggaaaaagt cggtaaagaaggtgtgattaccgttgaagaaggcaaaggccttgaagacgagcttgatgttgtagaaggcatgcagttcgatcgcggtt acttgtctccgtacttcatcaacaaccaagaaaaaatgaccgtagaaatggaaaatccattaattctattggttgataagaaaattgataac cttcaagagctgttgccaattcttgaaaacgtcgctaaatcaggtcgtccattattgatcgttgctgaagatgttgaaggccaagcactagc aacattggtagtaaacaacttgcgcggcacattcaaggttgcagcggttaaagcccctggttttggcgatcgtcgtaaagcgatgttgca agatcttgccatcttgacgggtggtcaggttatttctgaagagctagggatgtctttagaaactgcggatccttcttctttgggtacggcaagttggtgcgggttctgaaatggaaatgaaagaagaaagaccgtgttgacgatgcacttcatgcaactcgcgcagcggttgaagaag gtgttgttgcgggtggtggtgttgctttgattcgcgcactctcttcagtaaccgttgttggtgataacgaagatcaaaacgtcggtattgcat tgg cact tcgtgcgatggaagctcctatccgtcaaatcgcgggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcgggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcgggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcgggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcgggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcaggtgataacgcaggtgctgCagggGcagCggttgttgataaagtgaaatcgcaggtgataacgcagctgg cacagg tag ctttgg ttttaacgccag cacagg tgag tatggcg at at gattgcg at ggg tattttag acc ctgcaaa ag tcacgcag tagget at the context of thegttcatctctacaagccgcggcgtctatcgcaggtttgatgatcacaaccgaagccatggttgcggatgcgcctgttgaagaaggcgct ggtggtatgcctgatatgggcggcatggtggaatgggcggtatgcctggcatgatgtaatcactttgtgattcattgtcctgatctgcttaccgtGTCGACATATTCAAGATAAAGATGCCTTCACTGACATCAGTCACCAACAATC AATCAAACACCAATACCAATCGCAAAAACTCATAAAACTAGCCGATCACCAAAT CCCAAAAGCGTTCAAAAATGAAACGAGCACGTCACACAAAATCAATTTATACGC TAACGAACCAGGTCAAACTTATCGTTTTTTTGAGCACGTTTGTTCCACTAATGAAA GAGAAAAGTCGTTAATTCACTGGCTTTTTGGCGTATCCGCACCTTCACATAGAAAT TAGTAATGGCATGCTACTGGCCTTTAAAAAGAATCAGTTAATTGAAGAAACCTCG CTTATCTCAGCCATTACCGCTGTAGCCGAATTTGCGCTTATCCTCAGCCATGATTA AACTGACGCCAATTAATATAAGACATACTAATTAATAACTCCCTTAATTGAGAAG AATAATGAAAAACACACTCAAATCCTCATCACGTTTTAGTCTGAAACAACTCGGC ACCGGCGCTCTGATTATCTCCAGTTTGTTCTTCGGTGGTTGCACCACAACACACA AGATAATTTATACACAGGGGTTATGTCTCTTGCGAGAGACAGCGCTGGCCTAGAA GTTAAAACAGCCTCTGCCGGTGACGTCAATCTTACTTATATGGAACGCCAAGGCA GTGACAAAGATAATGCCGAAAGCGTTATTTTATTACACGGTTTCTCTGCTGATAA AGATAACTGGATTCTTTTTACCAAAGAATTCGATGAAAAATATCATGTTATCGCT GTCGATTTAGCGGGACATGGCGATTCAGAACAATTATTAACGACTGATTACGGTC TCATAAAACAAGCCGAGCGTTTAGATATCTTCTTATCTGGCTTAGGGGTTAACTC ATTTCACATCGCCGGTAATTCAATGGGGGGGGCTATCAGCGCAATCTACAGTTTG AGTCACCCAGAGAAAGTTAAAAGTCTTACATTGATCGATGCAGCAGGTGTCGATG GCGATACTGAAAGCGAATACTACAAAGTTTTGGCAGAAGGTAAGAATCCTTTAAT TGCAACTGATGAAGCAAGTTTTGAATACCGCATGGGTTTCACCATGACTCAGCCT

CCTTTCCTACCTTGGCCACTAAGACCTTCTTTATTACGTAAAACGCTAGCCCGTGC CGAGATCAATAACAAAATTTTTTCCGATATGCTGAAAACCAAAGAACGTTTAGGA ATGACTAACTTCAACAGAAAATTGAAGTGAAAATGGCTCAACATCCATTGCCAA CACTGATTATGTGGGGCAAAGAAGATCGCGTTCTTGACGTATCCGCAGCAGCGGC CTTCAAAAAAATAATTCCACAAGCAACTGTTCATATTTTTCCTGAAGTAGGCCAC CTACCTATGGTAGAAATTCCTAGTGAAAGCGCTAAAGTTTATGAAGAGTTTTTGT CCTCTATTAAATAAGAGCACATAATCATGACTGACTTATAAACAGCCAAGCATTT AAAATGCTTGGCTGTTTATTTTAATGGCCAAATTATTCAACGACCAAGCTCTGCG GTAAAATCGCAGTGGGTTTCTTGTTTTCATCAACAGCAACAAACGTGAAATACCC CGTAATCGCATTTTCTGATTATCAAAATACATACTTTCCACCAGCATATTAACTT CAACTTTTAAACTCGTCCGCCCTACCTCTATAACACTGGCAGTCAATTCGACAATG GTACCTGCGGGAACAGGATGCTTAAAATCGATTCGATCACTGCTGACGGTTACGA CATTGCAGTGCCACCGAATAACGTATCATGATGATTTGTTGTCTCTGGAAATACC GCTTTAGAAATAGTGGTTTTTGATACGCGCTTTCGCTGCGCAATAATATCTTCTCT GCTAAGAGTTGCGGATGGCATACATAAACTCGCTTGATTAAGATTAATAATAAAT AGTTAACAGTATATTGAACTGAGGGTCTGAAGAACTCTAATACCTCTGAAGAACT TTGAGGCCGCTAGAGAGAAAAGACCAGTGATAATATTTCATCTTGCCATGAGAGC AATAATACCAATATATTCATATATATATTCACACTACCCTTATCTCACTAGACTT CCCGCGCATAGGCGCAAACAATCAACGCAAGTTCACAATAAAGCGGTTCGCTGC AACACATGCCCTAGCGTCTAAAGTAGCACGCACAACACTGGCCAGTCGTACTAGC CCCTTTGCGATTCGTGCAGACGAGCAACAAGCGCTATTAAACTTACCTAAATTTC TAACCACCACCATTGGTTCTTTTCCACAAACTCAAAAACTCGTCAAATCCGCTTG CAATTTAAACGCGATGACATAGATCTAATCGATTATCAAACCCGCATTCAAGCGC TCATTAAAAACGCACCACTGGCAAGAAGTTCTACCTGCACTGACCAATATGCAAG CGGCGGCGAAGAGCTGCCTTTGATCGATCAAGAAGAAGAGGGAGCAGCAAAGAGG AAAACAATCAAAAAGAGGAGAGCAATCAAATAAAAACGAGTTATTGAGGATTTT **AATTTTAAAACAGGTATATTAATACCCTCTCTCGTAGTAAACAATGACTGTATTTA** CACAAAATAAATAGAGGTATACCATGTCAAACATCTGGTTTGAAGTACCAAAG ATTGAAGTATTAAACCGTCAAATGGAAAATACTGCCTGCAGCAACTTAGGCATTC AAATTACAGAAATTGGCGATGATTATATCACTGGCACAATGCCAGCAGATGCACG TACCTTCCAGCCAATGGGACTGATTCATGGCGGCTCAAATGTATTGCTGGCAGAA ACACTGGGCAGCATGGCAGCTAACTGCTGTATTAATTTGTCTCAAGAATATTGTG

Figure 9:

Amino acid sequence of the stabilized single ring mutant Glu461Ala/Ser463Ala/Val464Ala of Cpn60:

SEQ ID No 14: Cpn10 (nucleotides 458-751of Figure 10):

MKIRPLHDRIVVRRKEEETATAGGIILPGAAAEKPNQGVVISVGTGRILDNGSVQALA VNEGDVVVFGKYSGQNTIDIDGEELLILNESDIYGVLEA

SEQ ID No 15: Cpn60 (nucleotides 458-751 of Figure 10):

MAAKDVLFGDSARAKMLVGVNILADAVRVTLGPKGRNVVIEKSFGAPIITKDGVSV
AREIELKDKFENMGAQMVKEVASQANDQAGDGTTTATVLAQAIISEGLKSVAAGMN
PMDLKRGIDKATAAVVAAIKEQAQPCLDTKAIAQVGTISANADETVGRLIAEAMEKV
GKEGVITVEEGKGLEDELDVVEGMQFDRGYLSPYFINNQEKMTVEMENPLILLVDKK
IDNLQELLPILENVAKSGRPLLIVAEDVEGQALATLVVNNLRGTFKVAAVKAPGFGD
RRKAMLQDLAILTGGQVISEELGMSLETADPSSLGTASKVVIDKENTVIVDGAGTEAS
VNTRVDQIRAEIESSTSDYDIEKLQERVAKLAGGVAVIKVGAGSEMEMKEKKDRVD
DALHATRAAVEEGVVAGGGVALIRALSSVTVVGDNEDQNVGIALALRAMEAPIRQI
AGNAGAAGAAVVDKVKSGTGSFGFNASTGEYGDMIAMGILDPAKVTRSSLQAAASI
AGLMITTEAMVADAPVEEGAGGMPDMGGMGGMGGMPGMM

Figure 10:

SEQ ID No 16: DNA sequence of the stabilized single ring mutant Glu461Ala/Ser463Ala/Val464Ala:

In the DNA sequence:

Small letters – the Cpn10-Cpn60 coding fragment,

Big bold letters = introduced mutations

atcaaaaaatgcagcaaggacagattcctgcccaagaattagcagaaggtttcttgttagcactggccggcgctttattattaacgccgg gttttgtcactgatgcgctgggttttacattactcgtccccgcgacgcgtaaagcgttggtccataaggtgattgcatttattacccctcgcatgatgactgcaagcagctttcaagcgacgggtagttttcaggaaggctcgtttaaagatgtacattcgcacactgactcgcaaagca cgataaattaaccatctatttttcacagaggcaatttagcctttgtttaccttattgatcctaatacttgggatccaacagttggagagtctagcaaatgaaaatccgtccattacatgatcgtattgttgttcgccgtaaagaagaagagaccgcaactgcgggtggtattattttacc gggcgctgcggcagaaaaaccaaatcaaggtgttgttatctctgtgggtactggccgtattcttgataatggttcagtgcaagcgctggcggttaacgaaggcgatgttgtcgtttttggtaaatactcaggtcaaaatactatcgatatcgatggtgaagaattattgattttgaatgaaggtcgtaacgttgttatagaaaaatcatttggtgcaccgatcatcaccaaagatggtgtttctgttgcgcgtgaaatcgaattgaaagacaaattcgaaaacatgggcgcacagatggttaaggaagttgcttctcaagccaacgaccaagccggtgacggcacaacgacagc gact gtactagcacaggcgattatcagcgaaggcttgaaatctgttgcggctggcatgaatccaatggatcttaaacgtggtattgataaagcta ccgatgaaacggttggtcgtttaattgctgaagcgatggaaaaagtcggtaaagaaggtgtgattaccgttgaagaaggcaaaggcctt gaagacgagcttgatgttgtagaaggcatgcagttcgatcgcggttacttgtctccgtacttcatcaacaaccaagaaaaaatgaccgta cgt ccattatt gat cgt tg cag at gt tg aag gc caa gc act ag caa catt gg ta gaa acaa ct tg cg cg gc acatt caa gg tt gctggcgcaggtactgaagcaagcgttaatactcgtgttgaccagatccgtgctgaaatcgaaagctcgacttctgattacgacatcgaaaa gttacaagaacgcgttgctaagcttgcgggcggcgttgccgtgattaaggttggtgcgggttctgaaatggaaatgaaagaagaaa gaccgtgttgacgatgcacttcatgcaactcgcgcagcggttgaagaaggtgttgttgcgggtggtggtggttgtttgattcgcgcactct

gggtaacgcaggtgctgCagggGcagCggttgttgataaagtgaaatctggcacaggtagctttggttttaacgccagcacaggtg agtatggcgatatgattgcgatgggtattttagaccctgcaaaagtcacgcgttcatctctacaagccgcggcgtctatcgcaggtttgat gatcacaaccgaagccatggttgcggatgcgcctgttgaagaaggcgctggtggtatgcctgatatgggcggcatgggtggaatggg cggtatgcctggcatgatgtaatcactttgtgattcattgtcctgatctgcttaccgtgtaaaaagatcaggctcaaggctgctataaaaa agccgtatctttgatgatgttgtctttctgctgaaaacgacattcttggagtgcggctttttttgattttggtcataaaattcagaatattgtgta attttatgtaactagctggcctataatgttgagttcctctgggtggcatgatctcatggtacttcacttaagcctgattcactgcg gctttaacagtaaaataataacgcaacgtagaaacataataagcgtatggcattaatgaagacggctgcatttaattcagatc

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